OIE twinning project: Benefits for the candidate lab

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Presentation outline

- Candidate lab: Biopharma (Morocco)
- Twinning project: BT & AHS
- Parent lab: IAH Pirbright (UK)
- Technical benefits
- Scientific benefits
- GLP improvement
- Conclusion
- The way forward
SOCIETE DE PRODUCTIONS BIOLOGIQUES ET PHARMACEUTIQUES VETERINAIRES

- Creation in 1984
- Capital asset
  3.2 M USD under state control
- Investment
  12 M USD
- Human resources: 66 (27 technicians)
Candidate Lab: Biopharma

• Biopharma laboratory was founded in June 1985, its mission was to contribute, to preserve and enhance animal health assuring laboratory diagnostic and veterinary vaccine production.

• Biopharma has taken on diverse researches in the animal health sector for more than 25 years.

• At a national level, Biopharma is in charge of the diagnosis of contagious and exotic diseases as well conducting epidemiological surveys, and research projects to identify the main problems affecting animal health in the country.
Twinning Project

- African horse sickness (AHS) and Bluetongue (BT) are caused by viruses within the Orbivirus genus of the family Reoviridae.
- Since 1999 there have been widespread outbreaks of bluetongue in Europe and North Africa with many serotypes involved.
- In the last 3 years, there have been widespread outbreaks of AHS in sub-Saharan countries in Africa reaching the south borders of Morocco (Mauritania, Senegal).
- Project aims to enhance early detection and control of AHS and BT by improving laboratory diagnostic capacity, surveillance and technical expertise.
- Development of a centre of expertise for BT and AHS to assist countries in the North African region in their effort to combat the continuing and increasing threat of BT-AHS infection.
Parent lab: IAH Pirbright

- IAH Pirbright is OIE reference laboratory for BT & AHS and has a proven expertise in auditing, training and collaborating with laboratories from other countries.
- BT and AHS research and diagnostics have been carried out at Pirbright laboratory for over 30 years and the Laboratory holds one of the largest collections of BTV and ASHV strains in the world.
- Arbovirology program at Pirbright employs 23 full time staff and many PhD students who are all working on different aspects of entomology, molecular biology, immunology and diagnostics related to BT and AHS.
- The reference laboratory holds an annual 2 week training course specifically related to BTV diagnostics and molecular epidemiology. This is a practical course that involves both hands-on practical training in diagnostic methods as well as lectures given from the staff in their areas of expertise.
- The CRL also organizes an annual ring-trial for the European Community in which panels of sera and blood are sent around to labs in the EC.
Technical objectives

- Early detection and differentiation of AHS, BT viruses by applying conventional gel based PCR technique, real time PCR and nucleic acid sequencing
- Training for Biopharma laboratory staff
  - Virus identification and characterization
  - Improvement of serological techniques to OIE standards
  - Vector identification and surveillance
- Post training assessment
  Final workshop for dissemination of the information within regional labs in the country
- Establishment & management of a sample bank
Technical benefit 1

Technology transfer:

1. Early Diagnostic of the infection by real time PCR using commercial or home made kit

2. Serotyping by real time PCR:
   - Advantage: Fast and highly specific, results within hours instead of 7 days using serology
   - Adoption of a disease surveillance program based on sentinel animals checked periodically by rt-PCR

3. Virus isolation on insect cells
   - Simple and sensitive technique compared to egg isolation
Technical benefit 2

Providing specific reagents:

- Reference strains and of BTV: 24 serotypes
- Reference sera of BTV: 24 serotypes
- Reference strains of AHSV: 09 serotypes
- Reference sera of AHSV: 09 serotypes
- BTV Detection kit by ELISA
- BTV Detection PCR kit
Technical benefits 3

• Participation in the training course on diagnostic techniques for 5 Biopharma technical staff:
  o Virus isolation
  o Real time PCR
  o Antibody detection by ELISA
  o Serotyping by viral neutralization test
  o Introduction to virus sequencing
Technical benefit 4

• Dissemination of information:
  • Workshop on BT and AHS (2 days)
  • Rabat October 2010
  • 17 participants from regional labs, vet school and livestock Dept

✓ Presentation of the OIE and the twinning project
✓ World & regional situation of BT & AHS
✓ Technical presentation: molecular, serological diagnostic, surveillance and early warning
✓ Project perspectives
Assistance to improve “Good Laboratory Practices”

 Assistance in validation of diagnostic methods
 Strengthening quality assurance procedures
 Auditing the diagnostic lab by the IAH staff
 Participation in the annual ring-trial for the European Community labs
   – Genome detection by Real time PCR
   – Antibody detection by ELISA
   – Serotyping by viral neutralization test
Scientific benefit

- Access to scientific publications and journals in IAH Pirbright library
- Follow up epidemiological situation using molecular tools for risk assessment and early warning
- Exchange of biological materials:

BTV sequencing of all isolates from 2004 to 2010.

Evidence from full sequence analyses that MOR2009/09 is a reassortant strain (BTV-4R), containing sequences from previous BTV-1 and BTV-4 strains that spread in Morocco and neighbouring regions.

Detection of a new reassortant BTV1/4:

However both Seg-2 and Seg-6 (encoding outer capsid proteins VP2 and VP5) are from BTV-4, the BTV-4 serotype would be unchanged.
Similarity between BTV-4 2009 (MOR 2009/09) and previous strains of BTV 1 and 4 from Morocco, showing evidence of reassortment

<table>
<thead>
<tr>
<th>BTV-4 MOR 2004/02</th>
<th>BTV-4 Morocco 2009 (MOR 2009-09)</th>
<th>BTV-1 MOR 2006/06</th>
</tr>
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<tbody>
<tr>
<td>% similarity with MOR 2009-09</td>
<td>Genome segment Number</td>
<td>% similarity with MOR 2009-09</td>
</tr>
<tr>
<td>98.8%</td>
<td>1</td>
<td>99.8%</td>
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<tr>
<td>99.6%</td>
<td>2</td>
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<tr>
<td>94.1% (4MOR2009/10)</td>
<td>7</td>
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</table>

Source: P. Mertens
Publication of researches conducted during the period of the twinning:

**Experimental infection of camels with bluetongue virus**
Research in Veterinary Science (2010)

**Midge-transmitted bluetongue in domestic dogs**
C. A. L. OURA AND M. EL HARRAK
Epidemiol. Infect., Page 1 of 5. *f Cambridge University Press 2010*

**Development of RT-PCR assays for seven serotypes of epizootic haemorrhagic disease virus and their use to identify isolates from the Mediterranean region and North America**
Narender S. Maan, Sushila Maan, Kyriaki Nomikou, Mehdi El Harrak, Hagai Yadin, Hafsa Madani, Donna Johnson, Kadir Yesilbag, Carrie Batten, Simon J. Anthony and Peter P.C. Mertens
PLoS ONE 2011
Collaboration on other diseases

- **Peste des Petits Ruminants (PPR):**
  Validation of diagnostic technique by use of Ab ELISA detection kit based on HA protein

- **Sheep Pox:**
  Development of Ab detection Elisa kit

- **EHD:**
  Providing specific antiserum for EHD diagnostic and serotyping
  Experimental infection for Moroccan strain pathogenecity study
The OIE twinning project between Biopharma and IAH Pirbright was conducted successfully on two emerging diseases of great importance for Morocco and the Mediterranean region: BT and AHS.

This project enabled the laboratory to improve the scientific level of its technicians through a better knowledge of diseases, diagnostic techniques and epidemiological surveillance.

The project also supplied reagents, strains and reference serum required for the execution of diagnostic techniques.

Training of technicians and the parent lab assistance allowed technology transfer of diagnosis and monitoring.
CONCLUSION 2

Participation in the proficiency test organized by EC and the obtained results provide evidence that Biopharma laboratory has the required level to become an OIE regional RL. This collaboration was rewarded by scientific publications in well-known research journals.

Collaboration has been extended to other important diseases in the region, not included in the initial project.

The final workshop allowed the dissemination of scientific and technical information to regional laboratories and other institutions involved in BT/AHS control.
PERSPECTIVES

• Continuing collaborations ongoing research projects on BTV reservoir (dogs, camels), EHD pathogenecity.
• Regional dissemination of the information: Organization of a workshop on BT/AHS diagnostic & surveillance for North and Western Africa countries (Rabat 2011)
• Submitting candidature for OIE regional Reference Lab for BT/AHS
• Extend collaboration on other diseases of importance in the region (Sheep Pox and PPR).
Thank you